EXHIBIT 6k

brain gene transcription, transcription factor binding, and chromatin. For example, the discovery of NHIP (neuronal hypoxia inducible, placenta associated) through an epigenome-wide association in placenta identified a common genetic risk for ASD that was modified by prenatal vitamin use.

While epigenomic signatures are distinct between different genetic syndromic causes of ASD, bivalent chromatin and some convergent gene pathways are consistently epigenetically altered in both syndromic and idiopathic ASD, as well as some environmental exposures. Together, these epigenomic signatures hold promising clues towards improved early prediction and prevention of ASD as well as genes and gene pathways to target for pharmacological interventions. The author suggests that future advancements in single cell and multi-omic technologies, machine learning, as well as non-invasive screening of epigenomic signatures during pregnancy or newborn periods are expected to continue to impact the translatability of the recent discoveries in epigenomics to precision public health.

The study by Schmidt et al., examined pregnancy and environmental factors collected by maternal interviews as predictors of placental DNA methylation, including partially methylated domains (PMDs), an embryonic feature of the placental methylome.[480] The study analyzed DNA methylation data from MethylC-seq analysis of 47 placentas of children clinically diagnosed at 3 years with autism spectrum disorder (ASD) or typical development using standardized assessments. The data was examined in relation to various factors, including child's gestational age, birthweight, and diagnosis; maternal pre-pregnancy body mass index, smoking, education, parity, height, prenatal vitamin and folate intake; home ownership; pesticides professionally applied to lawns or gardens or inside homes, pet flea/tick pouches, collars, or soaps/shampoos used in the 3 months prior to or during pregnancy. The strongest and most robust associations were found between pesticides professionally applied outside the home and higher average methylation over PMDs and a reduced proportion of the genome in PMDs. The authors suggest that pesticide exposures could alter placental DNA methylation more than other factors.

The study by Dou et al examined the use of prenatal vitamins in the first month of pregnancy in relation to cord blood and placenta DNA methylation in two prospective pregnancy cohorts: the Early Autism Risk Longitudinal Investigation (EARLI) and Markers of Autism Risk Learning Early Signs (MARBLES) studies.[481] In the placenta, prenatal vitamin intake was marginally associated with lower mean array-wide DNA methylation in EARLI, and associated with lower mean array-wide DNA methylation in MARBLES. There was little consistency in the associations between prenatal vitamin intake and single DNA methylation site effect estimates across cohorts and tissues, with only a few overlapping sites with correlated effect estimates. However, the single DNA methylation sites with p-value < 0.01 were consistently enriched in neuronal developmental pathways. The authors conclude that their findings suggest that prenatal vitamin intake in the first month of pregnancy may be related to lower placental global DNA methylation and related to DNA methylation in brain-related pathways in both placenta and cord blood.

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[480] Schmidt et al. Self-reported pregnancy exposures and placental DNA methylation in the MARBLES prospective autism sibling study. Environ Epigenet. 2016 Dec;2(4):dvw024. doi: 10.1093/eep/dvw024. Epub 2016 Dec 1. PMID: 28781890; PMCID: PMC5538262.

[481] Dou et al. Prenatal vitamin intake in first month of pregnancy and DNA methylation in cord blood and placenta in two prospective cohorts. Epigenetics Chromatin. 2022 Aug 2;15(1):28. doi: 10.1186/s13072-022-00460-9. PMID: 35918756; PMCID: PMC9344645.

The study by Mordaunt reported differences in cord blood DNA methylation between children later diagnosed with autism spectrum disorder (ASD) and typically developing controls.[482] The study analyzed DNA methylation data from cord blood samples of 152 children clinically diagnosed at 3 years with ASD or typical development using standardized assessments. The data was examined in relation to numerous factors, including child's gestational age, birthweight, and diagnosis. At birth, prior to the diagnosis of ASD, a distinct DNA methylation signature was detected in cord blood over regulatory regions and genes relevant to early fetal neurodevelopment. Differential cord methylation in ASD supports the developmental and sex-biased etiology of ASD and provides novel insights into potential mechanisms underlying the disorder.

The article by Schroeder et al. discusses the use of placental methylome analysis as a potential tool for predictive ASD biomarkers in high-risk families.[483] The study used data from the MARBLES (Markers of Autism Risk in Babies: Learning Early Signs) prospective study and applied whole genome bisulfite sequencing to investigate human term placentas. The study found that while human placental methylomes have highly reproducible PMD and HMD locations, there is a greater variation between individuals in methylation levels over PMDs than HMDs due to both sampling and individual variability. In a comparison of methylation differences in placental samples from 24 ASD and 23 typically developing (TD) children, a HMD containing a putative fetal brain enhancer near DLL1 was found to reach genome-wide significance and was validated for significantly higher methylation in ASD by pyrosequencing. The study concludes that their results suggest that the placenta could be an informative surrogate tissue for predictive ASD biomarkers in high-risk families.

Convergent Pathways in ASD

The study examined the transcriptional differences between iPSC-derived cortical neurons from patients with idiopathic ASD and unaffected controls over a 135-day course of neuronal differentiation.[484] The study found that ASD-specific misregulation of genes was involved in neuronal differentiation, axon guidance, cell migration, DNA and RNA metabolism, and neural region patterning. Furthermore, functional analysis revealed defects in neuronal migration and electrophysiological activity, providing compelling support for the transcriptome analysis data. This study identified common processes altered in early neuronal development and corticogenesis that may contribute to ASD pathogenesis.

A similar study used organoid models of the human cerebral cortex to identify cell-type-specific developmental abnormalities that result from haploinsufficiency in three ASD risk genes—SUV420H1 (also known as KMT5B), ARID1B and CHD8—in multiple cell lines from different donors.[485] The authors reported that each of the three mutations confers asynchronous development of two main cortical neuronal lineages γ-aminobutyric-acid-releasing (GABAergic) neurons and deep-layer excitatory projection neurons but acts through largely distinct molecular pathways. Although these phenotypes are

[482] Mordaunt et al. Cord blood DNA methylome in newborns later diagnosed with autism spectrum disorder reflects early dysregulation of neurodevelopmental and X-linked genes. Genome Med. 2020 Oct 14;12(1):88. doi: 10.1186/s13073-020-00785-8. PMID: 33054850; PMCID: PMC7559201.

[483] Schroeder et al. Placental methylome analysis from a prospective autism study. Mol Autism. 2016 Dec 15;7:51. doi: 10.1186/s13229-016-0114-8. PMID: 28018572; PMCID: PMC5159983.

[484] DeRosa et al. Convergent Pathways in Idiopathic Autism Revealed by Time Course Transcriptomic Analysis of Patient-Derived Neurons. Sci Rep 8, 8423 (2018). https://doi.org/10.1038/s41598-018-26495-1

[485] Paulsen, B., Velasco, S., Kedaigle, A.J. et al. Autism genes converge on asynchronous development of shared neuron classes. Nature 602, 268–273 (2022). https://doi.org/10.1038/s41586-021-04358-6

consistent across cell lines, their expressivity is influenced by the individual genomic context, in a manner that is dependent on both the risk gene and the developmental defect. This study identified cell-type-specific neurodevelopmental abnormalities that are shared across ASD risk genes and are finely modulated by human genomic context, finding convergence in the neurobiological basis of how different risk genes contribute to ASD pathology.

EMBRYOLOGY AND TERATOLOGY

The science of embryology has a long and well-developed knowledge base pertaining to human gestation and fetal development. It also provides tools to scientists addressing questions of causation. As such, this discipline represents an important resource and discipline available to scientists addressing questions of causation.

For instance, embryologists have long since worked out the sequence and timing for the development of specific organs and organ systems in most species. Using this data and information, it is possible to know during which week of gestation a particular organ or system developed, and at what time during gestation its development has been completed. In broad terms, damage to organ systems caused by teratogenic or *in utero* insults can be often linked to specific gestational periods, termed critical windows of development (Figure 34). Of course, if no toxic insult occurred at this time that would argue against a chemical etiology for those malformations. On the other hand, where the time of the insult and the damaged organs match up, that can be important support for environmental causation.

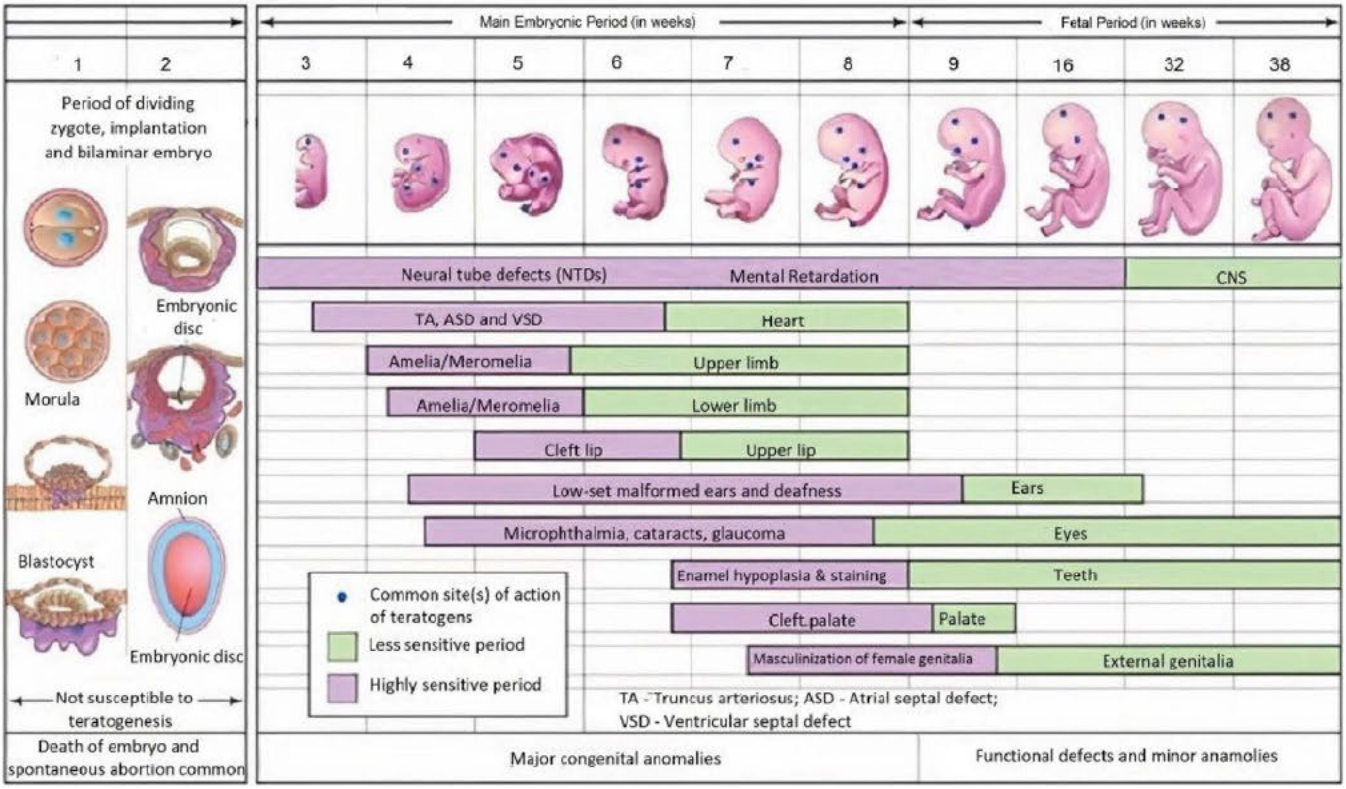

Figure 34. Critical Windows of Development. Depending on the nature of the toxic exposure, death or congenital malformations may result during the embryonic period. During the fetal period, the majority

of organ development is complete, so functional defects or minor anomalies are more likely from toxic exposures.[486]

Teratology is the branch of Embryology that focuses on environmental insults that have the capacity to damage embryological processes critical to the formation of healthy organs and systems. Teratology is the study of abnormal embryonic development. The founding principles of the modern study of teratogens were initially articulated by James G. Wilson (1973), co-founder of The Teratology Society, in his monograph *Environment and Birth Defects.*[487] These six principles remain important underpinnings of teratology, shepherding the study and knowledge of teratogens and their effects on developing organisms. These scientific principles continue to be employed when attempting to understand the potential of an environmental compound or pharmaceutical product to disrupt normal embryogenesis. These six principles are as follows:

- **Principle #1**: *Susceptibility to teratogenesis depends on the genotype of the conceptus and the manner in which this interacts with adverse environmental factors.* This principle speaks to the fact that there are genetically determined differences in susceptibility between individuals, with some being more sensitive to the teratogenic effects of a compound than others.

- **Principle #2**: *Susceptibility to teratogenesis varies with the developmental stage at the time of exposure to an adverse influence.* There are critical periods of susceptibility to agents and organ systems affected by these agents. This means that you cannot expect to induce cleft palate, for example, in a fetus if the exposure to the drug/compound takes place after the palate has formed. This principle applies to all of the major organs and structures of a developing embryo.

- **Principle #3**: *Teratogenic agents act in specific ways on developing cells and tissues to initiate sequences of abnormal developmental events.* This means that the compound disrupts normal development by interfering with specific physiological requirements of a developing organism. Teratogens act via specific mechanisms to cause birth defects. Unfortunately, we do not always know what these teratogenic mechanisms of action are, so that we cannot always develop effective interventions.

- **Principle #4**: *The access of adverse influences to developing tissues depends on the nature of the influence.* This means that there are several different factors that affect the ability of a teratogen to physically interact with a developing conceptus, such as the nature of the agent itself, route and degree of maternal exposure, rate of placental transfer and systemic absorption, and nature of the maternal and embryonic/fetal genotypes. Simply put, there are different exposure routes depending on whether the agent is a chemical, a virus, an X-ray or microwave, which influences the method by which the substance in question enters the mother's body to gain access to the developing embryo.

- **Principle #5**: *There are four manifestations of deviant development (Death, Malformation, Growth Retardation and Functional Defect).* This means that a compound could be considered to be a teratogen even if it did not cause structural malformations in an embryo. A substance that

[486] From Moore and Persaud. The Developing Human. 10th edition. Elsevier Health Sciences. ISBN: 9780323313483
[487] Wilson, James Grave. Environment and Birth Defects. New York: Academic Press, 1973.

causes the embryo to die, for whatever reasons, is a teratogen. Same is true for compounds that cause growth restrictions or adversely impacts mental development, even in the absence of structural malformations.

- **Principle #6**: *Manifestations of deviant development increase in frequency and degree as dosage increases from the No Observable Adverse Effect Level (NOAEL) to* a *dose producing 100% Lethality (LD100).* This means that as you increase the concentration of the drug exposure, you might expect to find a higher prevalence of malformed infants and that the severity of the malformations might also be expected to increase in severity with increasing dosage.

The field of teratology captured the public's attention in the early 1960s, when the morning sickness medication, Thalidomide, was determined to be the cause of thousands of children being born with limb reductions, known as phocomelia, as well as other congenital malformations.[488] This may have been the most significant event in the development and implementation of drug testing regulations that are intended to protect expectant women and their unborn children from the teratogenicity of clinically available medications.

Teratologists are typically well-trained embryologists who readily appreciate the fact that a causation analysis cannot be limited to, or founded upon, the absence of data respecting a particular unusual or rare outcome. From an embryological perspective, numbers of rare outcomes or the presence of multiple anomalies are usually compelling evidence for an environmental etiology. Finally, and of particular importance, embryological studies have clearly established that the fetus is far more vulnerable and thus far more susceptible to toxic insult and injury than are adult human beings. Accordingly, in assessing exposure levels, it must be appreciated that levels thought to be safe for adult exposures, will not necessarily be safe to the developing fetus.

This important methodological insight flows from the fact that, especially in the early weeks of gestation, many different organs and organ systems are being created simultaneously in highly interdependent and dynamic processes. Gestation begins with a single undifferentiated cell and over time these cells begin to differentiate as the fetus evolves from a cluster of identical cells to an ever more complex entity with a multitude of organs, and tissues.

[488] Friedman JM. ABCDXXX: The obscenity of postmarketing surveillance for teratogenic effects. Birth Defects Res A Clin Mol Teratol. 2012 Aug;94(8):670-6. doi: 10.1002/bdra.23043. Epub 2012 Jul 11. PMID: 22786781.

RULING OUT ALTERNATIVE EXPLANATIONS

I have reviewed the report prepared by ChemRisk. Regarding background information, they included a reference to a report by the Minnesota Department of Health (MDH). It should be noted that the reference doses used in that study are based on background exposures to PAP and APAP, not on pharmaceutical exposures to APAP. As indicated in **Acetaminophen and Aniline**, the pharmaceutical intake of APAP results in an approximately 1000-fold increase in exposure levels. This report, viewed in this context of pharmaceutical dosing does not present a 1700-fold margin of safety, only a relatively small margin of safety that is approximately 1.7 to 2.4 times therapeutic pharmaceutical exposures. This is similar to the margin of safety between APAP recommended dosages (1g x 4 = 4g/day) and the potential for toxicity at intakes ≥ 150 mg/kg (~7.5 g/day in adults). In this example, doubling the recommended dose (2g x 4 = 8g), would also exceed the margin of safety for APAP and liver damage.

It is important to understand that the dose used by the MDH was based on background environmental intake, not pharmaceutical intake. I will present the MDH exposure data compared to pharmaceutical exposures. The derivations of the MDH reference doses (RfDs) ranged from 0.093 to 0.28 mg/kg/day. A recommended human dose (1000 mg), is a human reference dose of 16.7mg/Kg, based on a 60Kg human. This single dose is 180 to 60 times higher than the MDH RfD, and a recommended daily dosage, once every six hours (1g x 4) produces an APAP dosage of 66.8mg/Kg/day, and this is 720 to 240 times the RfD.

The information presented by ChemRisk looks very different with this pharmaceutical dose in mind, as ChemRisk quotes the MDH report,

> "the overall weight-of-evidence suggests that acetaminophen is not a developmental toxicant in humans" and that "[e]xperimental animal studies do not suggest increased malformations from therapeutic use of acetaminophen during pregnancy" (MDH, 2015: p. 5). Additionally, it was noted that "[n]o effects on pregnancy or offspring were reported in several laboratory animal studies at human equivalent doses up to over 500 times higher than the acute and short-term RfDs" while "a continuous breeding animal study, effects on reduced fertility and reproduction were observed at human equivalent dose 800 times higher than the acute and short-term RfDs" (MDH, 2015: p. 5). Finally, it was noted that "[a]cetaminophen is not considered to be a neurotoxicant based on lack of secondary observations in animal studies" and that "[i]n laboratory animals, clinical neurotoxicity symptoms were reported only at very high doses over 1,700 times higher than the RfDs" (MDH, 2015: p. 5).

No effects at over 500 times the RfDs, would be a good margin of safety, but this safety is relative to environmental exposures, such as municipal drinking water. This exposure is within the range of a daily pharmaceutical intake, as the daily dosage of 4g/day for APAP results in 720 to 240 times the RfD. Likewise, the continuous breeding animal study reported reduced fertility and reproduction at 800X times the acute and short-term RfDs, which is actual only 1.1-3.3 times a daily "therapeutic" intake. Much of the ChemRisk report presents the data in this fashion, which could be rather misleading without the appropriate context.

As another example of this type of presentation, the ChemRisk analysis incorrectly combine rat and mouse dosing and compare directly to human dosages. Specifically, ChemRisk compares rat and mouse dosing

to human dosages without converting to HEDs, human equivalent dosages, based on mg/m^2. The absence of this surface area conversion, which is used to account for metabolic differences between species, results in ChemRink incorrectly suggesting that dosages used in animal studies are too high to be relevant for human exposures or are overwhelmingly expected to cause hepatoxicity. The majority of mouse and rat studies are within therapeutic dosages, as indicated in Table 6 and Table 9. Dosages used in mice and rats, respectively, are reported and include experimentally determined therapeutic antinociceptive dosages (ED_{50}) for mice and rats, and estimated HEDs below the tables, reported and calculated as previously indicated, see Table 1 and Human Equivalent Dosages.

I have considered the following explanations, as presented by Alwan et al.[489] (2021) in response to Bauer et al (2021).[490] Alwan proposes, (1) "failure to account for confounding, and elements of bias that make interpretation of the data challenging", (2) "confusion with other analgesic drugs", (3) "Residual confounding from shared co-morbidities and genetic and environmental", (4) "migraine or fever", or (5) "underlying genetic susceptibility to neurodevelopmental and psychiatric conditions." Confounding was addressed, and multiple study designs addressed confounding with different approaches, yet the individual studies and meta-analyses still detected statistically significant associations. Regarding confusion, other analgesics have not reported the same risks, so such confusion should bias towards the null, not away. Regarding genes and environment, specific genetic risk factors are expected to be identified showing increased risks for individuals or populations; this is expected and consistent with Wilson's Principles. Regarding migraine or fever, other medications also pose risks during pregnancy, and these confounders were controlled for in studies individually and in meta-analyses. Regarding a familial risk or underlying genetic susceptibility, there are specific genes that can cause ASD and ADHD, but that does not negate the population burden and risk of exposing pregnant women to APAP, that increased the prevalence in that population above background. Pregnancy itself also increases the risk of APAP toxicity, and having underlying genetic risk factors is expected to increase the severity of disease or the incidence in APAP exposed population.

Bauer presents the following graphic, which summarizes animal data in "a call for precautionary action."

[489] Alwan et al. Paracetamol use in pregnancy - caution over causal inference from available data. Nat Rev Endocrinol. 2022 Mar;18(3):190. doi: 10.1038/s41574-021-00606-x. PMID: 34907341.

[490] Bauer et al. Reply to 'Paracetamol use in pregnancy - caution over causal inference from available data'; 'Handle with care - interpretation, synthesis and dissemination of data on paracetamol in pregnancy'. Nat Rev Endocrinol. 2022 Mar;18(3):192. doi: 10.1038/s41574-021-00610-1. PMID: 34907342.

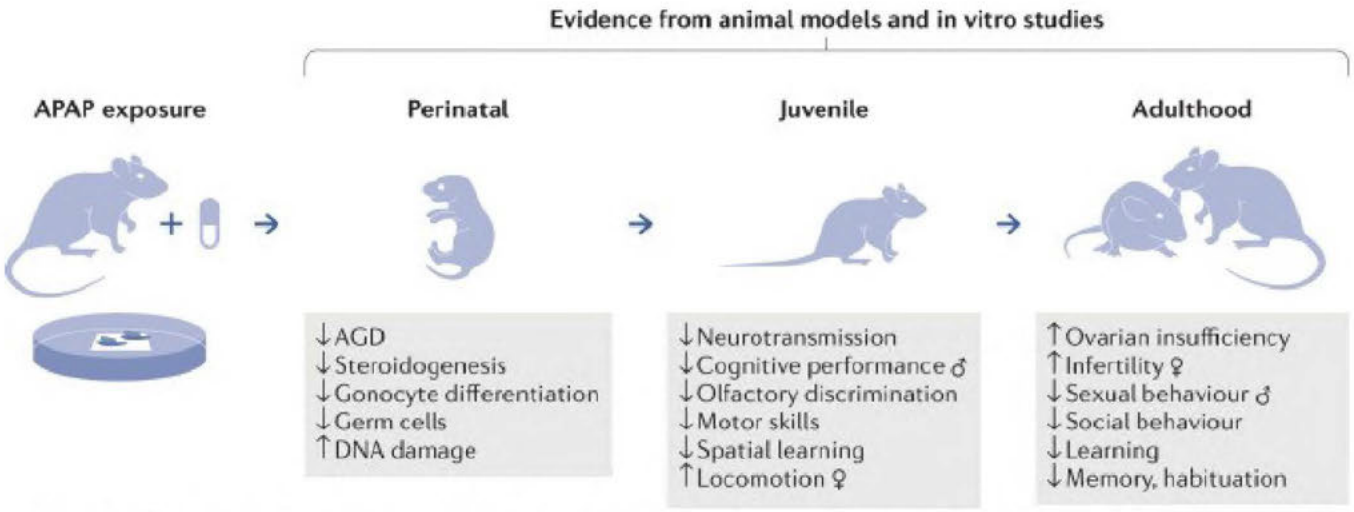

Figure 35. Evidence of Acetaminophen (APAP) Disruption of Reproductive and Neurological Development from Animal Studies (from Bauer et al. 2022).

As reviewed above, there is "**clear evidence" of reproductive, developmental, and neurodevelopmental toxicity.** The NTP reports there is "equivocal evidence" of carcinogenicity based on leukemia in female rats, which is consistent with oxidative damage and DNA damage, known to be produced by the APAP metabolite NAPQI. I find there is **"some evidence"** of carcinogenicity based on the similar cancers found in female rats by NTP (1993), and the increased incidence of leukemia in human taking APAP, as a function of dose and duration.

Regarding reproductive or developmental toxicity:

- Clear evidence of reproductive toxicity is demonstrated by a dose-related effect on fertility or fecundity, or by changes in multiple interrelated reproductive parameters of sufficient magnitude that by weight of evidence implies a compromise in reproductive function.
- Clear evidence of developmental toxicity is demonstrated by data that indicate a dose-related effect on one or more of its four elements (embryo-fetal death, structural malformations, growth retardation or functional deficits) that is not secondary to overt maternal toxicity.

Regarding carcinogenicity:

- Some evidence of carcinogenic activity is demonstrated by studies that are interpreted as showing a chemical-related increased incidence of neoplasms (malignant, benign, or combined) in which the strength of the response is less than that required for clear evidence.

Bauer presents the following graphic, which summarize human data in "a call for precautionary action."

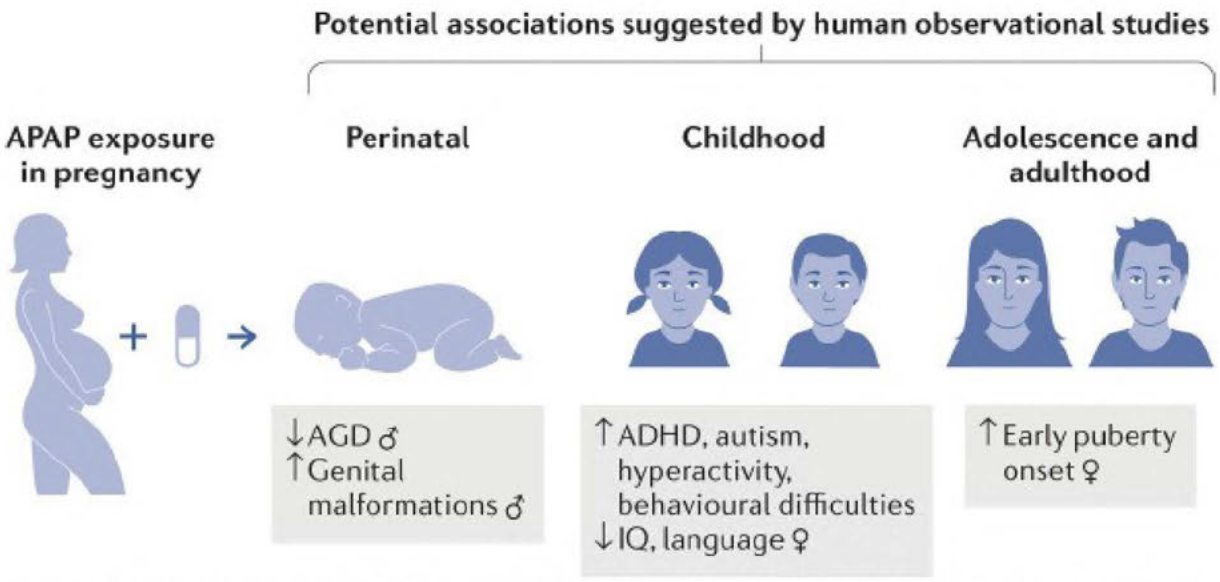

Figure 36. Associations between Prenatal APAP Exposure, Reproductive and Neurobehavioural Development Suggested from Observational Human Studies (From Bauer et al. 2022).

Bauer presents the following regarding use of medications during pregnancy,

> These findings suggest that many people view APAP as conveying negligible risk, instead of being a 'true medication' with potential adverse effects.[491]

Much like the evidence initially rejected by doctors that smoking could cause lung cancer, the historic usage of APAP has built a false belief of safety. Regulatory and research toxicity studies, based on both clinical and animal testing, indicate an increase in risk with frequent or continuous APAP usage. This false perception of safety is demonstrated by APAP being considered safe, while covertly being the primary cause of acute liver failure in the United States. While historic usage creates claims of safety to an unknowing public, as did smoking, these false beliefs must yield to factual evidence and scientific data. The evidence is clear, APAP taken during pregnancy, at therapeutic dosages, is sufficient to cause and increase the incidence of, as a function of duration and timing, reproductive, developmental, and neurodevelopmental toxicity.

[491] Bauer et al. Reply to 'Paracetamol use in pregnancy - caution over causal inference from available data'; 'Handle with care - interpretation, synthesis and dissemination of data on paracetamol in pregnancy'. Nat Rev Endocrinol. 2022 Mar;18(3):192. doi: 10.1038/s41574-021-00610-1. PMID: 34907342.

THE CAUSAL RELATIONSHIP BETWEEN ACETAMINOPHEN AND NEURODEVELOPENTAL TOXICITY

A systematic review of the genetic, reproductive, developmental, and neurodevelopmental toxicity literature indicates that APAP and metabolites thereof are associated with reproductive damage, developmental toxicity, teratogenicity, neurodevelopmental toxicity, and neurobehavioral and neurodevelopmental delays. Association, however, does not alone prove causation and further analysis is necessary. Sir Bradford Hill proposed a set of criteria to determine causation between exposure and outcome. These criteria are accepted in the scientific community as a reliable and viable method of determining the potential existence of causality. Thus, I next review each of the Bradford Hill criteria in terms of their importance for establishing that there is a causal relationship between acetaminophen and neurodevelopmental toxicity.

1. Strength of Association:
   The greater the magnitude of the association between an environmental exposure and an adverse outcome, the more likely it is that the exposure caused the outcome. In general, an odds ratio between 1 and 2 is deemed low, a ratio from 2 to 6 is deemed moderate, and a ratio above 6 is deemed high. A low strength of association, however, is not by itself sufficient to reject a cause-and-effect hypothesis. As Hill stated: "We must not be too ready to dismiss a cause-and-effect hypothesis merely on the grounds that the observed association appears to be slight," because "[t]here are many occasions in medicine when this is in truth so."[492] We must carefully assess whether other factors (confounders) may be responsible for an observed low or more moderate strength association. And if high quality studies reasonably account for identified confounders and still show an association (particularly a dose-response association), then the mere potential that some other unidentified factor is responsible for the excess risk is not a basis for rejecting the causation hypothesis. As Hill explained, "If we cannot detect it or reasonably infer a specific [alternative cause], then in such circumstances I think we are reasonably entitled to reject the vague contention of the armchair critic 'you can't prove it, there may be such a feature'."

   Numerous studies are cited that report an increased risk for neurodevelopmental toxicity, with ADHD and ASD as common endpoints, and APAP exposure. The magnitude of this risk is low in the majority of studies (1<OR<2), but moderate risk (OR>2) is reported with higher acetaminophen exposures. Specifically, there are reported risks of ADHD with APAP in meconium resulting in an OR 2.43 (95% CI, 1.41-4.21); with a dose-response doubling of exposure increasing the odds of ADHD by 10% (OR, 1.10; 95% CI, 1.02-1.19), and when grouped based on high levels of APAP detected in meconium, the odds of ADHD were more than 4-fold, OR 4.10 (95% CI, 2.41-6.95).[493] Measurements of unmetabolized APAP in umbilical cord blood at >50th percentile were also associated with higher odds of ADHD diagnosis (aOR: 2.10, 95% CI, 1.43- 3.11, $p < 0.001$).[494] While fever is proposed as a confounder of these reported associations, termed confounding by

[492] Hill, A.B. The environment and disease: association or causation? (1965) Proc R Soc Med. 1965 May;58(5):295-300. PMID: 14283879; PMCID: PMC1898525.
[493] Baker et al. Association of Prenatal Acetaminophen Exposure Measured in Meconium With Adverse Birth Outcomes in a Canadian Birth Cohort. Front Pediatr. 2022 Apr 5;10:828089. doi: 10.3389/fped.2022.828089. PMID: 35450103; PMCID: PMC9017809.
[494] Anand et al. Perinatal Acetaminophen Exposure and Childhood Attention-Deficit/Hyperactivity Disorder (ADHD): Exploring the Role of Umbilical Cord Plasma Metabolites in Oxidative Stress Pathways. Brain Sci. 2021 Sep 30;11(10):1302. doi: 10.3390/brainsci11101302. PMID: 34679367; PMCID: PMC8533963.

indication, there is a lack of evidence to support that other medications used for the same indications (fever), such as ibuprofen or aspirin, are associated with ADHD or ASD risks, which means that the presence of fever does not explain the increased risk associated with exposure to APAP. In addition, there are factors that bias studies towards the null, which makes the association weaker. Overall, meta-analyses report statistically significant associations between acetaminophen exposure and ASD[495] and ADHD[496] risk. While some individual studies have not supported specific associations, the totality of data is consistent with "clear evidence of developmental toxicity" as stated by the National Toxicology Program (NTP), "Clear evidence of developmental toxicity is demonstrated by data that indicate a dose-related effect[497] on one or more of its four elements (embryo-fetal death, structural malformations, growth retardation or functional deficits) that is not secondary to overt maternal toxicity."

Moreover, animal studies remove indications and other confounders, and as supported by numerous well controlled animal studies, APAP has been shown to cause neurodevelopmental toxicity and impaired learning and social behaviors consistent with ASD and ADHD at clinically relevant therapeutic dosages. These reports meet the criterion of strength of association.

2. Consistency of the Evidence:
This criterion assesses whether the observed association has been "repeatedly observed by different persons, in different places, circumstances, and times," which then allows us to "justifiably infer that the association is not due to some constant error or fallacy that permeates every inquiry." [498]

The association between fetal exposure to APAP and ASD/ADHD has been reported in epidemiological studies performed by several different laboratories working independently and publishing in different peer-reviewed journals. The association has been observed in Spanish, Brazilian, Swedish, Norwegian, English, and Danish cohorts, each independently reporting neurodevelopmental impacts, including increased ASD, ADHD, and associated behaviors such as language delays.

The epidemiological studies have also used differing techniques and methodologies, including prospective cohort studies, followed longitudinally without foreknowledge of the outcomes.

The meta-analyses combine data from multiple cohort studies, indicating a consistent pattern of association between acetaminophen exposure and ASD and ADHD risk across different populations. While some studies have not supported specific associations, the totality of data is consistent with "Clear evidence of developmental toxicity." As stated by the National Toxicology Program (NTP), "Clear evidence of developmental toxicity is demonstrated by data that indicate a

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[495] Alemany et al. Prenatal and postnatal exposure to acetaminophen in relation to autism spectrum and attention-deficit and hyperactivity symptoms in childhood: Meta-analysis in six European population-based cohorts. Eur J Epidemiol. 2021 Oct;36(10):993-1004. doi: 10.1007/s10654-021-00754-4. Epub 2021 May 28. PMID: 34046850; PMCID: PMC8542535.
[496] Ricci et al. In utero acetaminophen exposure and child neurodevelopmental outcomes: Systematic review and meta-analysis. Paediatr Perinat Epidemiol. 2023 Mar 20. doi: 10.1111/ppe.12963. Epub ahead of print. PMID: 36939050.
[497] The term "dose-related" describes any dose-response relationship, recognizing that the test article-related responses for some endpoints may be non-monotonic due to saturation of exposure or effect, overlapping dose-response behaviors, changes in immunologic manifestations at different dose levels or other phenomena.
[498] Hill, A.B. The environment and disease: association or causation? (1965) Proc R Soc Med. 1965 May;58(5):295-300. PMID: 14283879; PMCID: PMC1898525.

dose-related effect[499] on one or more of its four elements (embryo-fetal death, structural malformations, growth retardation or functional deficits) that is not secondary to overt maternal toxicity." In addition, well-controlled animal studies from at least [X] different laboratories have independently demonstrated "clear evidence" that perinatal APAP exposure to mice and rats, at doses equivalent to human therapeutic dosages, caused neurodevelopmental toxicity, changes in the brain, and impaired learning and socialization. The consistency criterion is met.

3. Specificity:
A high degree of specificity exists to the extent exposure to one factor (and no other) causes one disease (and no other). As a practical matter, high specificity can be detected when a rare environmental event is associated with a rare disease. The usage of APAP is common to a majority (>50%) of human pregnancies and the frequency of ASD (~2.2%) and ADHD (~10%) are increasing in the USA. The association between APAP exposure and ASD or ADHD is not specific since APAP exposure is common and can cause other toxicities, including hepatotoxicity. ASD and ADHD can also be caused by other exposures, such as *in utero* valproic acid or mercury exposure. When controlled for in animal studies or adjusted for in epidemiological studies, specific interactions are present. The specificity criterion is not fully met.

4. Temporality:
Epidemiological and animal studies demonstrated that exposure preceded outcome. Studies have shown that the effect of APAP is dependent on the timing of exposure in relation to specific developmental processes. The exposure to acetaminophen (during pregnancy) precedes the outcome (development of ASD or ADHD) temporally, supporting a temporal relationship. The temporality criterion is met.

5. Biologic Gradient (dose-response):
The meta-regression analyses indicate that the association between APAP exposure and ASD risk increased with the child's age at follow-up and the mean duration of exposure. This supports a biological gradient, as a longer duration of exposure and older age were associated with a higher risk of ADHD. The biological gradient criterion is met for ASD.

Meta-analyses did not explicitly report a dose-response relationship between APAP exposure levels and the risk of ADHD. Two studies looked at APAP in meconium, one supporting a dose-response interaction between APAP and ADHD. Another study looked at cord blood and supported a dose-response interaction. The biological gradient criterion is supported for ADHD.

Dose-response is also reported throughout testing for reproductive and developmental toxicity. APAP and the metabolite NAPQI increase oxidative damage in a dose dependent manner, which in turn produces genetic, reproductive, developmental, and neurodevelopmental toxicity. The biological gradient criterion for neurodevelopmental toxicity is supported.

6. Biologic Plausibility:
There is biological plausibility for the interaction, as APAP can cross the placental barrier and affect

[499] The term "dose-related" describes any dose-response relationship, recognizing that the test article-related responses for some endpoints may be non-monotonic due to saturation of exposure or effect, overlapping dose-response behaviors, changes in immunologic manifestations at different dose levels, or other phenomena.

fetal neurodevelopment. The published adverse outcome pathway (OECD 20) also supports a causal-plausible interaction between APAP and oxidative stress and neurodevelopmental adverse outcomes. The brain is particularly vulnerable to oxidative stress. When GSH is depleted, free radicals—including NAPQI and other endogenous radicals that normally rely on GSH buffering—cause damage that disrupts normal metabolic, cellular, and neurodevelopment of brain tissues in multiple different ways. NAPQI can bind molecules within cells and damage or disrupt the normal functions of proteins, lipids, DNA, and other components of the cell. Oxidative stress induced damage disrupts the differentiation of stem cells into neurons, proliferation of cells, migration of neural cells, and mitochondrial functioning, and can cause overt toxicity and cell death.

In addition to generating oxidative stress, APAP operates on a parallel mechanism by disrupting endocannabinoid, prostaglandin, and serotonergic signaling. The enzyme FAAH, which produces the APAP metabolite AM404, is expressed in neurons and throughout the central nervous system. APAP and its metabolite AM404 disrupt the endocannabinoid system by increasing levels of anandamide, the primary CB1 agonist, and by activating both CB1 and CB2 receptors. APAP also inhibits synthesis of prostaglandins. The effect of APAP on the endocannabinoid system and prostaglandin synthesis is sufficient to relieve pain and fever in an adult. But the normal operation of the endocannabinoid system and prostaglandins are essential for normal brain development. Endocannabinoids regulate the number and division rate of stem cells that differentiate into neural cells, the type of neuronal cell (neuron or glia) produced from neural stem cells, axon growth, and axon positioning. Prostaglandins are used to signal the formation of dendritic spines, and to influence arborization and pruning of connections. AM404 is also a potent activator of TRPV1, which can activate an inflammatory response in the brain. APAP also has indirect effects on serotonergic signaling, and such signaling facilitates neurogenesis, cell migration, synaptogenesis, and synaptic plasticity. The neurodevelopmental impact of acetaminophen on ADHD and ASD risk are biologically plausible and supported.

7. Coherence:

This criterion assesses whether the cause-and effect interpretation of the data is consistent with or would "seriously conflict with the generally known facts of the natural history and biology of the disease."[500] Hill identifies as examples whether causation is consistent with any sex differences in the disease, and whether we can isolate the particular factor in the environmental exposure that can cause the outcome in a laboratory setting (e.g. a chemical in cigarette smoke that causes cancer on the skin of laboratory animals). This is fulfilled by the epidemiological, global adverse reporting, and animal studies on pre-, peri- and post-natal APAP exposures.

A causal relationship between APAP and neurodevelopmental harm is consistent with the expected timing and results of exposure. For APAP to cause neurodevelopmental harm, we would expect that exposure during key developmental windows would produce harm. Animal studies demonstrate that exposure to APAP at therapeutic doses during the Brain Growth Spurt (PD3 or PD10, but not on PD19) results in later-life changes in the brain and impaired learning and socialization. If APAP exposure at therapeutic doses can generate sufficient oxidative stress to result in neurodevelopmental toxicity, then we expect that it would also cause parallel damage on

[500] Hill, A.B. The environment and disease: association or causation? (1965) Proc R Soc Med. 1965 May;58(5):295-300. PMID: 14283879; PMCID: PMC1898525.

reproductive development and DNA oxidation damage. Human and animal studies have demonstrated that perinatal exposure to APAP is associated with reproductive toxicity and DNA oxidative damage. While adverse outcomes are seen across numerous body systems, they are linked by oxidative damage, consistent with the published APAP AOP (see, Figure 11. Oxidative Stress and Developmental Impairment of Learning and Memory).

APAP as a causative agent is consistent with sex-linked differences in ASD and ADHD. The male to female ratio in ASD has been recently estimated as 4:1.[501] The estimates of the male to female ratio for ADHD range from 3:1 to 10:1.[502] The degree of symptoms is also reported to vary between males and females. For example, males with ASD have been reported to exhibit more repetitive behaviors than females, and males with ADHD are reported to greater inattention, hyperactivity/impulsivity, and greater total ADHD symptoms than females with ADHD.[503] Consistent with these outcomes, exposure to APAP in animal studies has resulted in sex-linked differences. The association between APAP exposure and ASD and ADHD is coherent with the existing knowledge of oxidative stress and potential for hepatotoxicity and neurotoxicity with APAP exposures. The coherence criterion is met.

8. Experimental Evidence:
Randomized controlled trials for developmental toxicity, teratogenicity, or neurodevelopmental toxicity are not conducted in pregnant women for ethical reasons. The available experimental evidence from animal models consistently demonstrates dose-responsive reproductive, developmental, and neurodevelopmental toxicity with pre-, peri- and post-natal APAP exposures. This criterion is supported with available experimental evidence.

9. Analogy:
The presence of an analogous drug-disease causal relationship supports a causal inference. There are analogies with other substances that are known to have neurodevelopmentally toxic effects during pregnancy, including Δ9-THC, mercury, and valproic acid, supporting a common oxidative stress mechanism for mercury, like APAP, and chemical-pharmaceutical causes of ADHD and ASD.

I reviewed above the evidence showing that Δ9-THC from cannabis has a common mode of action with the APAP metabolite AM404. As reviewed, AM404 inhibits the uptake of anandamide, and Δ9-THC and anandamide can both act as agonists for CB1 and CB2 receptors, meaning they can bind to and activate these receptors. Altering cannabinoid signaling during development produces a shared outcome, neurodevelopmental toxicity, including ASD and ADHD.

Another analogy is mercury. As explained above, the OECD prepared an Adverse Outcome Pathway analysis on a set of chemicals that bind to sulfhydryl (thiol)-group proteins. The results

[501] Centers for Disease Control and Prevention (CDC) Prevalence of autism spectrum disorder among children aged 8 years—autism and developmental disabilities monitoring network, 11 Sites-United States, 2014 (2018, April 27). Retrieved from: https://www.cdc.gov/mmwr/volumes/67/ss/ss6706a1.htm. 10.15585/mmwr.mm6745a7

[502] Biederman J, Mick E, Faraone SV, Braaten E, Doyle A, Spencer T, et al. Influence of gender on attention deficit hyperactivity disorder in children referred to a psychiatric clinic. *Am J Psychiatr* (2002) 159:36–42. 10.1176/appi.ajp.159.1.36

[503] Mahendiran, T. et al., Sex Differences in Social Adaptive Function in Autism Spectrum Disorder and Attention-Deficit Hyperactivity Disorder (2019) Front. Psychiatry 10:607, DOI: 10.3389/ fpsyt.2019.00607

were published in December 2022 as AOP 20. Both mercury and APAP are reported to cause oxidative stress. The analysis concluded that exposure to mercury causes "oxidative stress" sufficient to "cause cellular injury and death" that disrupts "the establishment of neuronal connections and networks" and can "lead to functional impairment in learning and memory." In addition, two studies found that children living in close proximity to industrial power plant sources of mercury had significantly higher prevalence of autism.[504], [505] It has also been reported that children with autism exposed to mercury showed significantly decreased level of GSH, and exposure to mercury has been also reported in some cases of autism.[506] Oxidative damage during development produces a shared outcome, neurodevelopmental toxicity, including ASD and ADHD. Meta-analyses found a significant association between higher mercury levels in various tissues (blood, RBC, and brain) and ASD.[507] This suggests a positive correlation between mercury exposure and ASD risk, meeting the criterion of strength of association. The association between mercury exposure during pregnancy and ASD is not specific, as mercury exposure has also been associated with other adverse health effects and neurodevelopmental disorders. Thus, the specificity criterion is not fully met. Available meta-analyses do not provide detailed information on the timing of mercury exposure during pregnancy and the subsequent development of ASD in offspring. The temporality criterion cannot be fully evaluated based on the available information. The meta-analyses do not explicitly report a dose-response relationship between mercury exposure levels and the risk of ASD. Further information on the dose-response relationship is necessary to assess the biological gradient criterion. The plausibility of a causal relationship between mercury exposure during pregnancy and ASD is supported by the known neurotoxic properties of mercury. However, additional research is needed to elucidate the specific mechanisms through which mercury may contribute to the development of ASD. The association between mercury exposure during pregnancy and the development of ASD is coherent with the known neurotoxic effects of mercury. However, further coherence with other scientific knowledge and research is required for a more comprehensive assessment. Controlled experiments involving deliberate mercury exposure during pregnancy are not feasible due to ethical considerations. Therefore, the available evidence relies primarily on animal studies and observational studies, including case-control designs, as seen in meta-analyses. Based on the information provided in meta-analyses and the application of the Bradford Hill Criteria, there is evidence supporting an analogous causal relationship between mercury exposure during pregnancy and ASD.

Another analogous causal relationship is found in valproic acid. Several studies have demonstrated an association between valproic acid exposure during pregnancy and an increased risk of ASD. Valproic acid exposure during pregnancy has been linked to an increased risk of various neurodevelopmental disorders, including ASD. Although not specific to ASD alone, the fact that

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[504] Windham GC, Zhang L, Gunier R, Croen LA, Grether JK: Autism spectrum disorders in relation to distribution of hazardous air pollutants in the San Francisco bay area (2006) Environ Health Perspect 2006; 114: 1438–1444.

[505] Palmer RF, Blanchard S, Wood R: Proximity to point sources of environmental mercury release as a predictor of autism prevalence (2009) Health Place; 15: 18–24.

[506] Mutter J, Naumann J, Schneider R, Walach H, Haley B: Mercury and autism: accelerating evidence? (2005) Neuro Endocrinol Lett 26: 439–446.

[507] Jafari et al. The association between mercury levels and autism spectrum disorders: A systematic review and meta-analysis. J Trace Elem Med Biol. 2017 Dec;44:289-297. doi: 10.1016/j.jtemb.2017.09.002. Epub 2017 Sep 4. PMID: 28965590.; Sulaiman et al. Exposure to Aluminum, Cadmium, and Mercury and Autism Spectrum Disorder in Children: A Systematic Review and Meta-Analysis. Chem Res Toxicol. 2020 Nov 16;33(11):2699-2718. doi: 10.1021/acs.chemrestox.0c00167. Epub 2020 Oct 12. Erratum in: Chem Res Toxicol. 2021 Jun 21;34(6):1693. PMID: 32990432.

valproic acid has been consistently associated with neurodevelopmental effects strengthens the case for a potential causal relationship. The exposure to valproic acid occurs during pregnancy, and the subsequent diagnosis of ASD in the offspring is assessed later in life. This temporal sequence supports a potential causal relationship between the exposure and the outcome. Evidence suggests that there might be a dose-response relationship between the level of valproic acid exposure during pregnancy and the risk of ASD. Higher levels of exposure or longer duration of exposure may be associated with a greater risk of ASD. Valproic acid is known to affect neurodevelopment and has been associated with various adverse effects on the developing brain. Animal studies and in vitro research support the plausibility of valproic acid's potential role in the development of ASD. The association between valproic acid exposure during pregnancy and an increased risk of ASD is coherent with our current understanding of the neurodevelopmental effects of valproic acid and the etiology of ASD. As an analogy, there is also substantial evidence suggesting a causal relationship between valproic acid exposure during pregnancy and an increased risk of ASD.

The FDA-approved label for Depakene states, "Although the available studies have methodological limitations, the weight of the evidence supports a causal association between valproate exposure in utero and subsequent adverse effects on neurodevelopment, including increases in autism spectrum disorders and attention deficit/hyperactivity disorder (ADHD)."[508]

I find that APAP carries a number of risks, including those that manifest as the primary cause of acute liver failure in the United States. There is risk of encephalopathy, both proximal (rare) and secondary to hepatic damage, with a relatively limited margin (< 2X MRHD) of safety. There are also reproductive, developmental, and neurodevelopmental risks, as presented in this structured review and report, as supported by "clear evidence" in animal studies and "clear evidence" in available human meta-analyses examining the risk of *in utero* APAP exposures.

[508] New Drug Application (NDA): 018081, Company: ABBVIE, 05/19/2020, SUPPL-71, Labeling-Package Insert, DEPAKENE, VALPROIC ACID, https://www.accessdata.fda.gov/drugsatfda_docs/label/2020/018081s071,018082s054lbl.pdf

CONCLUSIONS

Within a reasonable degree of scientific certainty, the undersigned opines:

1. A therapeutic dose of Acetaminophen (APAP) produces >1000X increase in APAP exposures over environmental background exposures to APAP (from environmental aniline).

2. Acetaminophen produces oxidative damage, reproductive, developmental, and neurodevelopmental toxicity at clinically relevant therapeutic dosages, including neurodevelopmental disorders, such as autism spectrum disorder (ASD) and attention deficit hyperactivity disorder (ADHD).

3. Therapeutic dosages of APAP taken by pregnant woman are sufficient to cause neurotoxicity, neurodevelopmental disorder, ASD, and ADHD in exposed offspring.

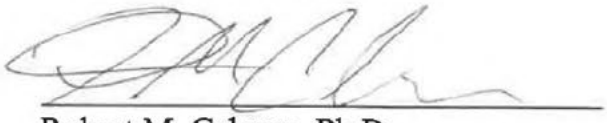
4. There exists no reasonable alternative cause or explanation for these injuries which contradict or undermine the general causation opinions expressed above.

I expressly reserve the right to amend or supplement this report and to read, review and comment upon any reports prepared by Defendants' experts.

The foregoing opinions are substantively identical to the opinions stated in my June 16, 2023, report. All opinions offered herein are held to a reasonable degree of scientific certainty.

Dated: June 22, 2023

Respectfully submitted,



Robert M. Cabrera, Ph.D.